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Figure 1A

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Figure 1B

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Figure 1D



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Figure 1F

V P R P R P G T E A T I T G L E P G T E V T I V V I A L K N N O K S E P L I G GETCOCTCORDODDDCTGETGECOCGAGGCTACTATTACTGGCCTGGAACCGGGAACCAATATACATTTATGTCATTCCCTGAGAAATAATCAGAAGACCCTGATTCC GTATGACACTOGAMATOGTATTCAGCTTCCTGCCACTTCTGGTCACCAACCCAGTGTTGGCAACAAATGATTTGAGGAACATGGTTTTAGGGGAGCACAAGGGCACAACGGCAC PIREPRETE TO BEALSOIIIS WAP FOOTSEVILS CHP 32 SATAGE VATVG NSVNEGLNOPTO OSCFOPTTVS NYAVG TOTGEGIOLP GISGOOPS V GOOMIFEEN GFRRIPPILAT THE RESESTABLE OF LOFGSONFRCOSSRUCHONGVNY A SE A SOMBSCICIONORGEFRCOPHENICIOO DE LA COLOR DE LA OCAGABOCA TANGGTT COCCEANGAGGETT GT TACCGT COCCANGE CANGGANG COCAGAGGAN TACCGT COT TOCAGATT TOCAFT AT TOCAFT TOCAF aga i gag toogaaqgaat i gigacag tig toogag tegettaggetti togaag tog i gataa togaatig togaatig togaatig togaatig CAGATTOGAGAAGTODDAQCGACAAAATODDCAGATGATGAGCTDCACATGTCTTODCAACOGAAAAQAGAAFTCAAGTGTGAOCCTCATGAODCAACGTGTACGATGA V G T D E E P L O F R V P G T S T S A T L T O L T R G A T V W I I V E A L <u>2</u> 8 R Š 20 ğ ž ş وه الحال الله مرحول الله يكومون الله الومون الله الله الواده Ş i 28

Figure 1G

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Figure 1H

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Figure 2A

Palr 2

Pair 1

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4 ACACAACCAAACAIGAACAAIACCICCIICGGICCAAAAIIGACGCIIICAIIIGGACIICGACIICICI GTTTGTACTTGTTATGGAGGAAGCCGAGGTTTTAACTGCGAAAGTAAACCTGAAGCT - 3'

A 714

GAAGAGACTTGCTTTGACAAGTACACTGGGAACACTTACCGAGTGGGTGACACTTATGAGCGTCTAAA GAACGAAACTGTTCATGTGACCCTTGTGAATGGCTCACCCACTGTGAATACTCGCAG - 5'

Figure 2B

Pair 5

01 1

Pair 6

S-- ATCGCCAACGCTGCCATGAAGGGGGTCAGTCCTACCAGATTGGTGACACCTGGAGGAGACCACATGAGACT - 3'
GACATGGTAGCGTTGCGACGGTACTTCCCCCAGTCAGGATGGTCTAACCACTGTGGACCTCCTGGTGTACTCTGACCACAA 112

Pale 7

. S GGTGGTTACATGTTAGAGTGTGTGTGTTTGGTAATGGAAAAGGAGAATGGACCTGCAAGCCCATAGCTGAG - 3' **TGTACAATCTCACACACACAGAACCATTACCTTTTCCTCTTACCTGGACGTTCGGGTATCGACTCCTAG** 113

DOKET " TAGENGO

11/62 Figure 3

Synthesis of F.N. Gene (14 - 472 b.p.)

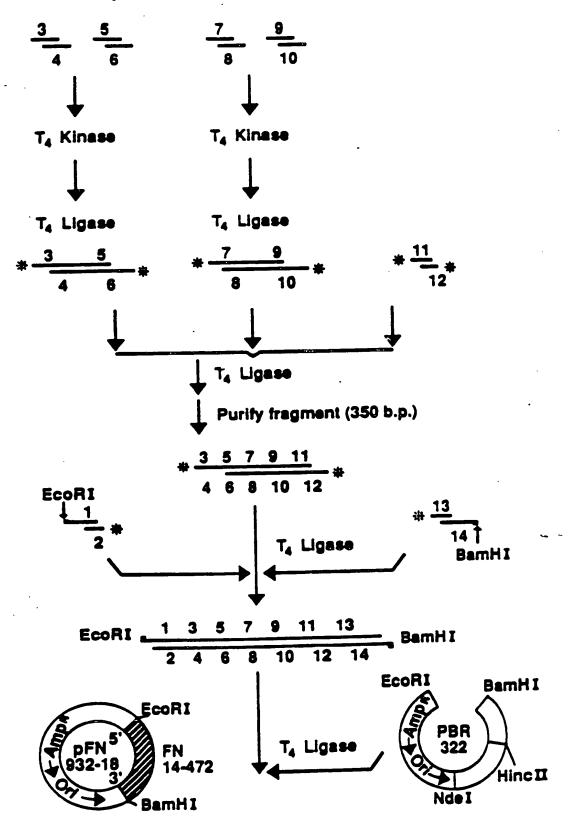
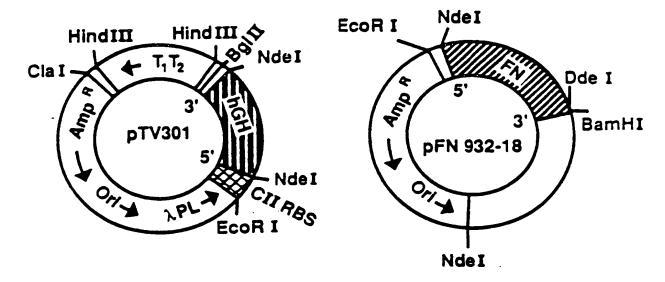
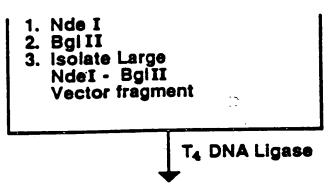
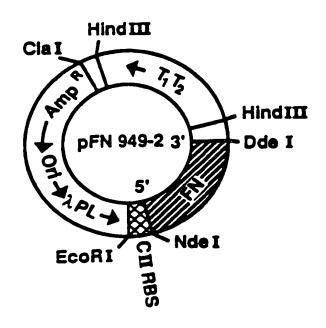


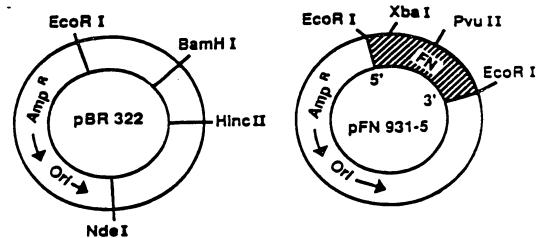
Figure 4

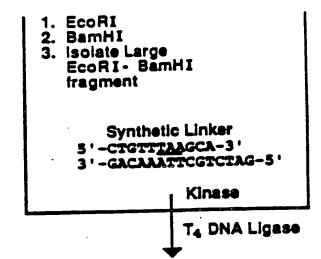




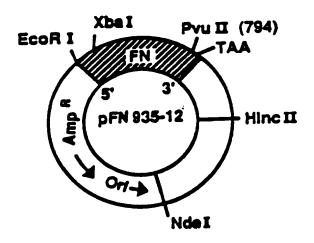
- 1. Nde I
- 2. BamH I
- 3. Isolate
 NdeI BamHI
 FN fragment

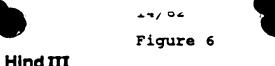


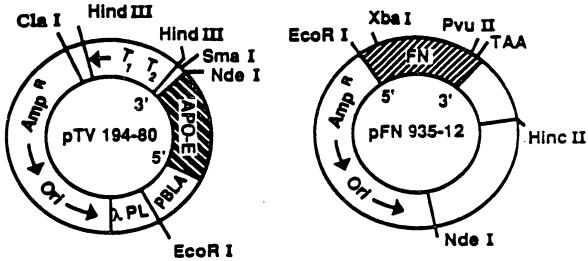




- 1. EcoRI 2. Pvu II 3. Isolate Large EcoRI - PVUII FN fragment



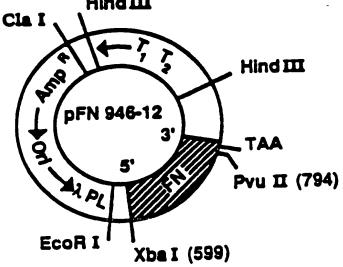




- 1. EcoRI
- 3. Isolate large EcoRI SmaI fragment

- 1. EcoRI 2. Hinc II
- 3. Isolate EcoRI - Hinc II FN fragment

T. DNA Ligase HInd III



Xba I (599)

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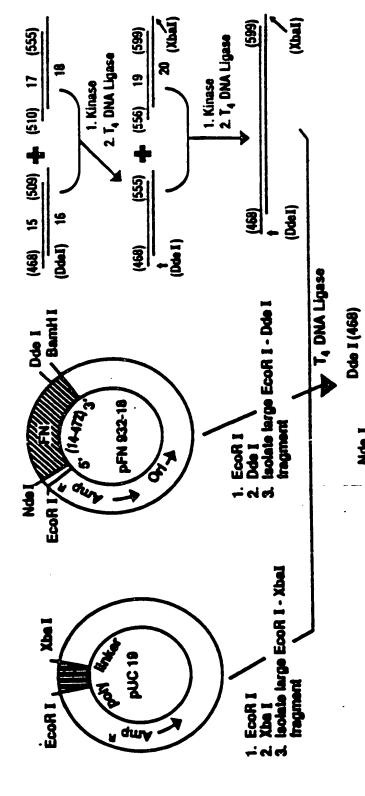
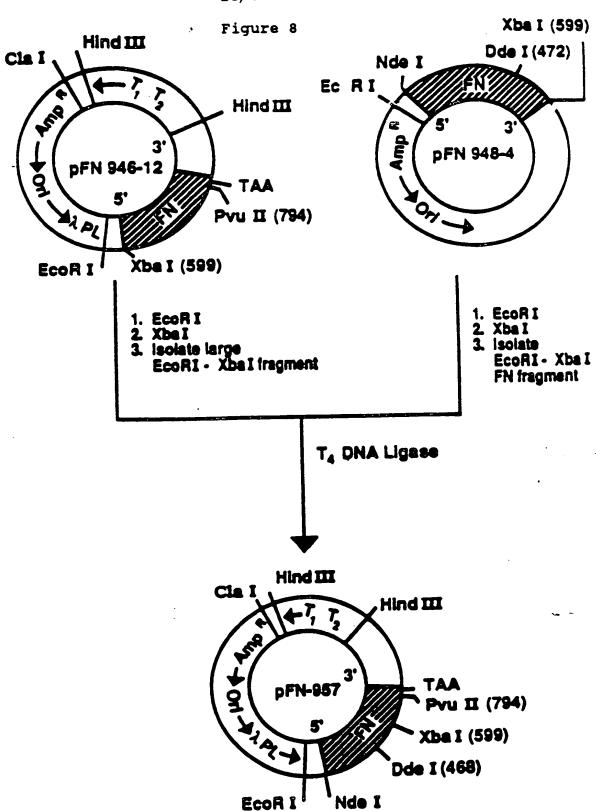


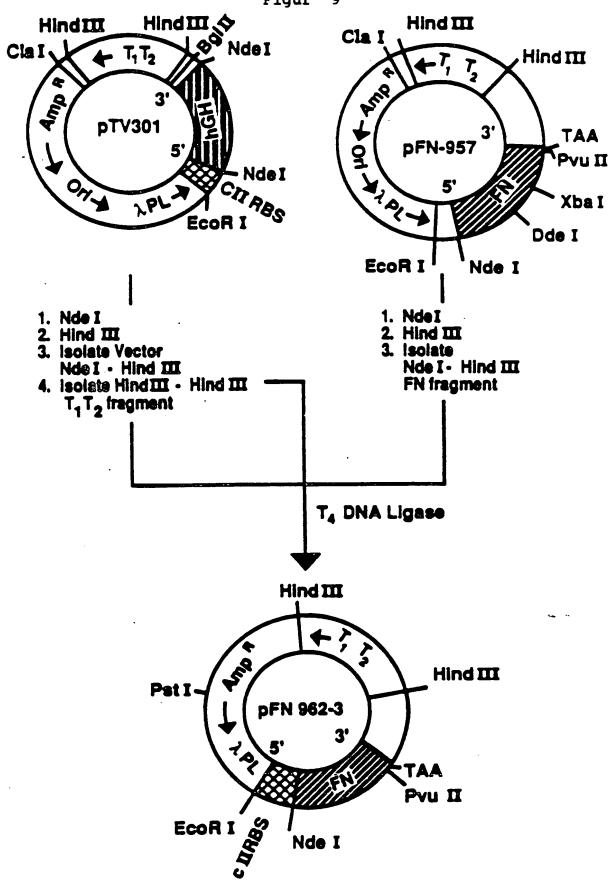
Figure 7

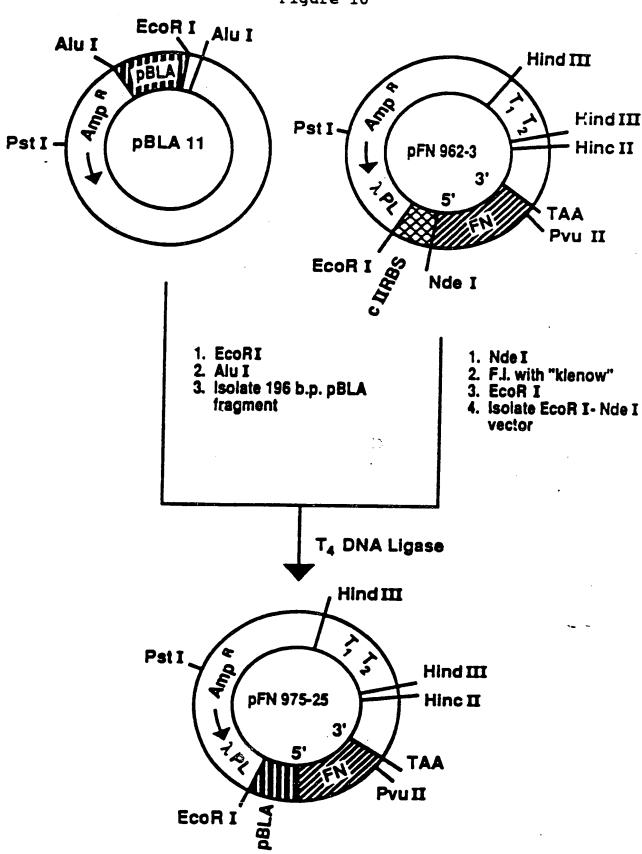
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Figur 9





19/62 Figure 11

REDUCED (+MZ)

NON-REDUCED (-ME)

p-31kD

M.WT. Markers

After Q-S

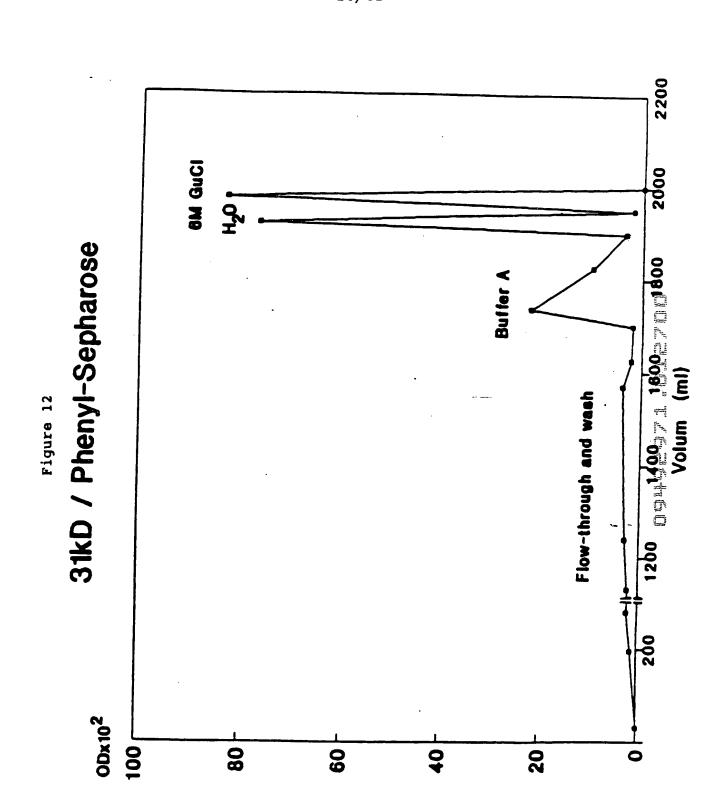
Extracted Pellet Refolded (CSH/CSSG) After Phenyl-S p-31kD

After Q-S

Refolded (CSH/CSSC)

After Phenyl-S

30kD →



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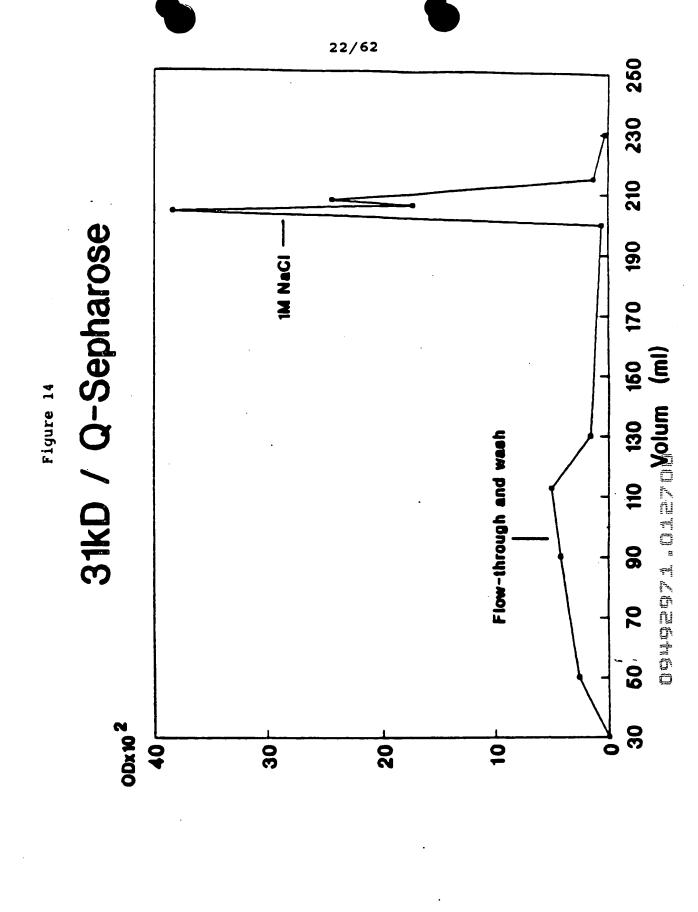
12

Figure 13 31kD/Heparin-Sepharose

ODX 10

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Mixture of rec. and plasmatic "31kD" FBD **10kD** FPLC - Superose 12 37kD *150kD ODx10² 8 8 10 (7)

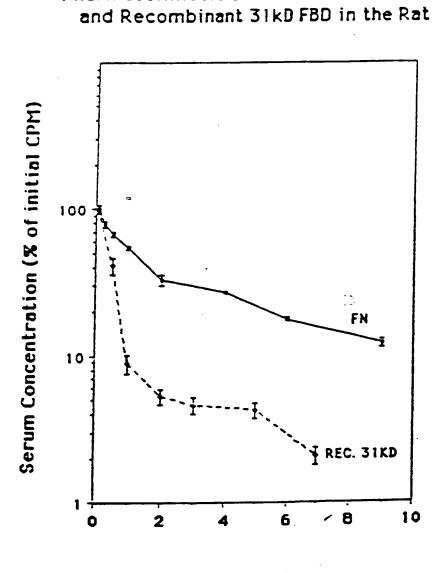
Figure 15

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Figure 16

Pharmacokinetic Behavior of Fibronectin



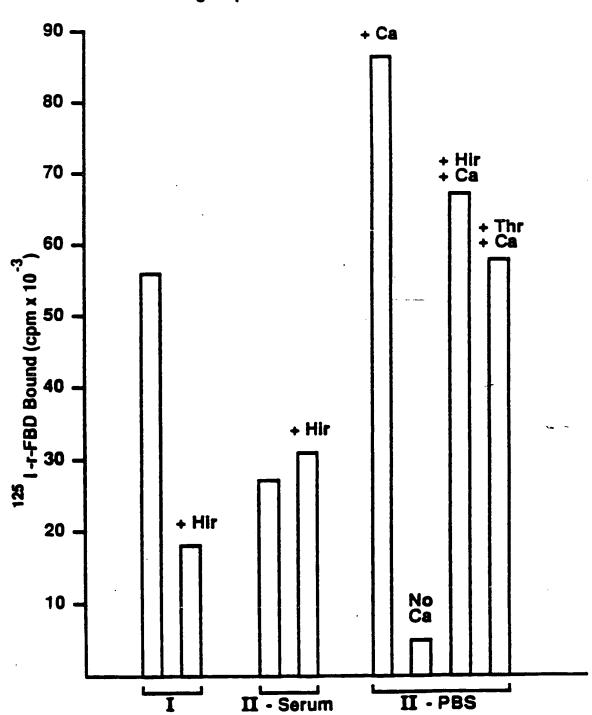
Time (h)

DO 4 O E 7 Z . D X E 7 D II

Binding of ¹²⁵ I -FBD to Fibrin; Effect of Thrombin and Ca⁺⁺ ions.

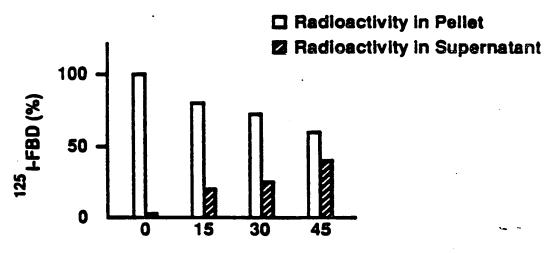
I = Binding while clot formation

 Π = Binding to preformed Fibrin ciot



26/62 Figure 18

Release of ¹²⁵ I-FBD from Fibrin Clot by Plasmin.



Time of incubation with Plasmin (min)

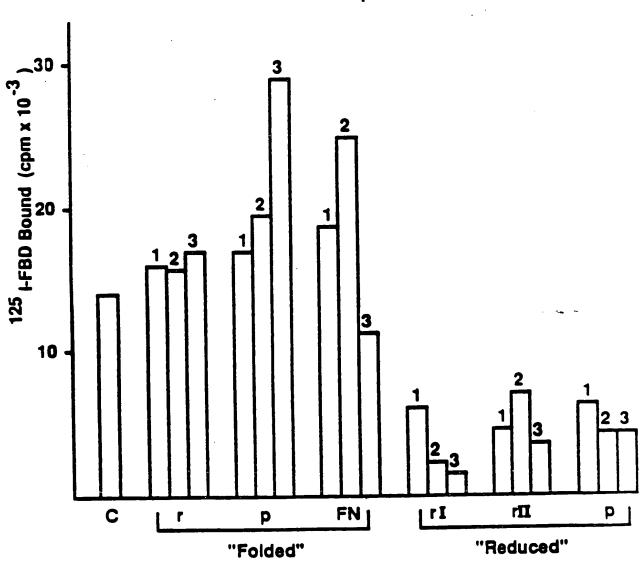
27/62 Figure 19

Binding of 125 I-FBD during clot formation (Reaction I); Effect of unlabelled FBD and related molecules.



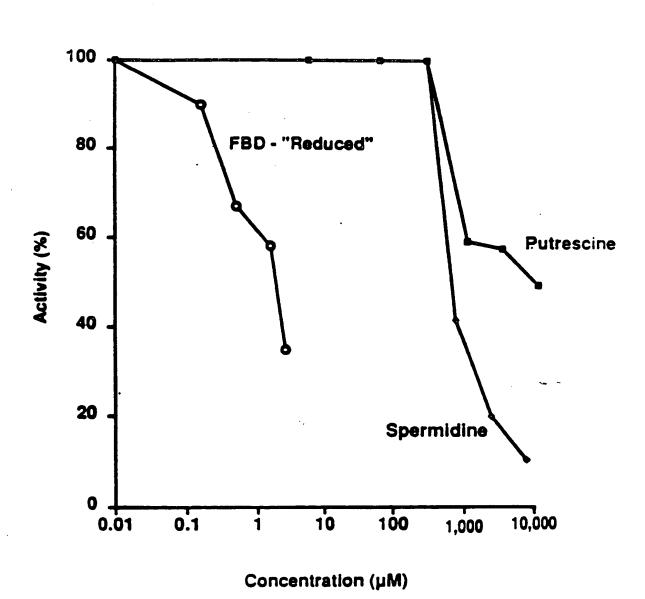
2 - 1.0 µM " "

3 - 3.0 µM "



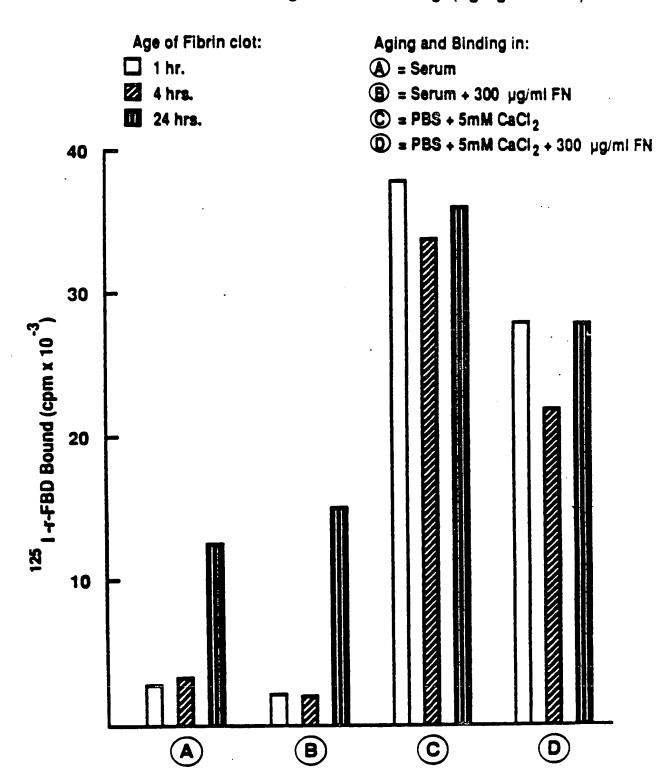
28/62 Figure 20

Binding of 125 I-FBD to Fibrin (Reaction II); Effect of Transgiutaminase Inhibitors.





Binding of 125 I -FBD to preformed Fibrin clot (Reaction II); Effect of Fibrin clot Age on the Binding (aging at 37° C).





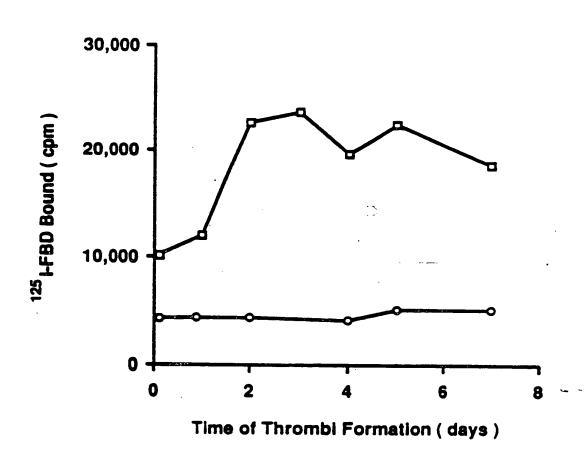


30/62

Figure 22

Binding of FBD to "Naive" Thrombi

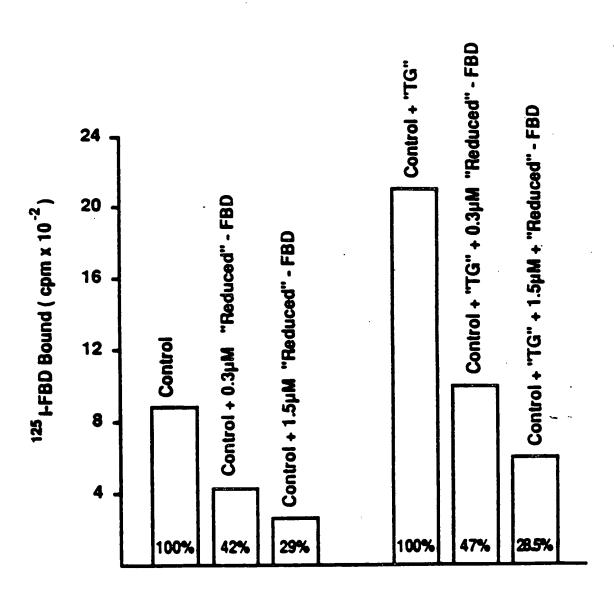
--> Preformed ---- Newly - formed





Figur 23

Binding of FBD to Fibrin (Reaction I); Effect of Transglutaminase ("TG") and "Reduced" - FBD





Binding of ¹²⁵I-FBD to ECM, Effect of Ligand Concentration and Thrombin

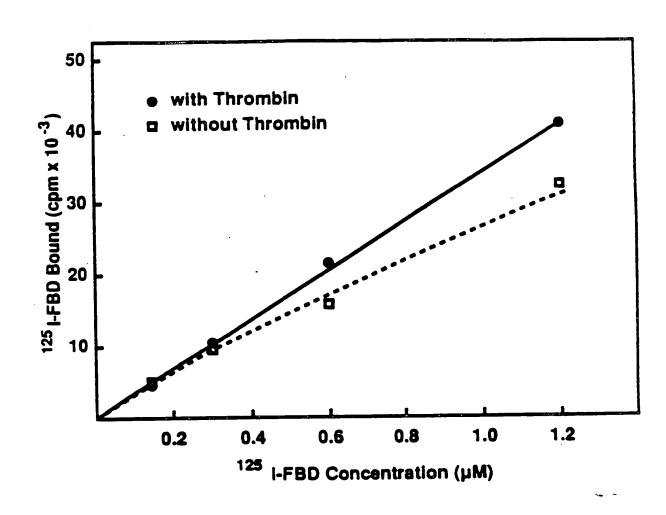
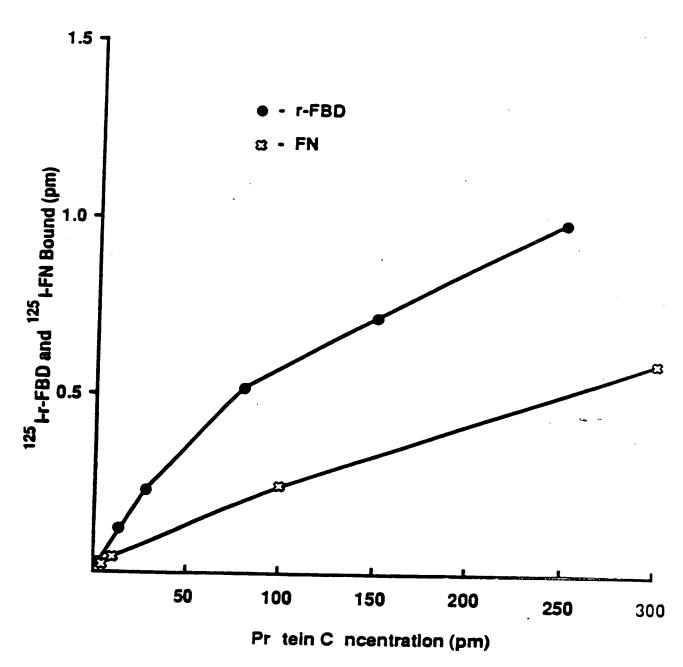
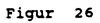


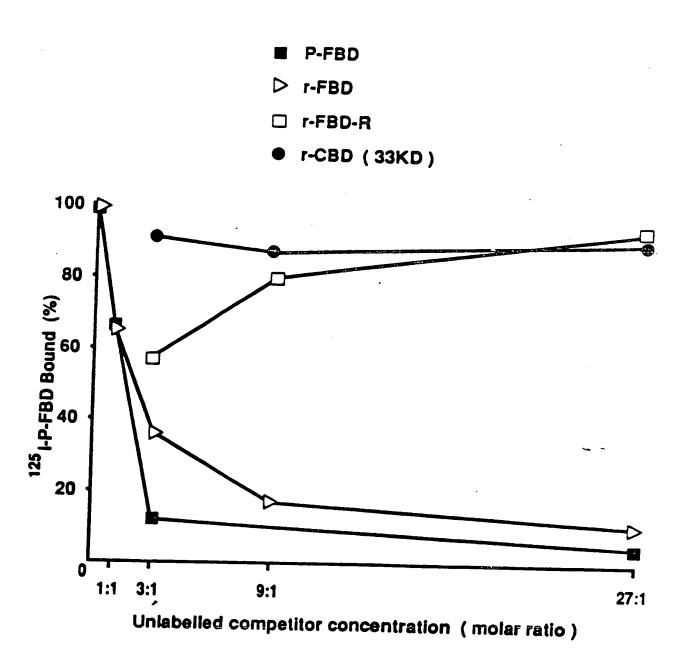
Figure 25

Binding of 125 I-(FN, r-FBD) to S. aureus





Binding of 125 I-FBD to S. aureus; Competition with "Folded" and "Reduced" forms.



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Figure 27

Binding of S. aureus to Immobilized FN.

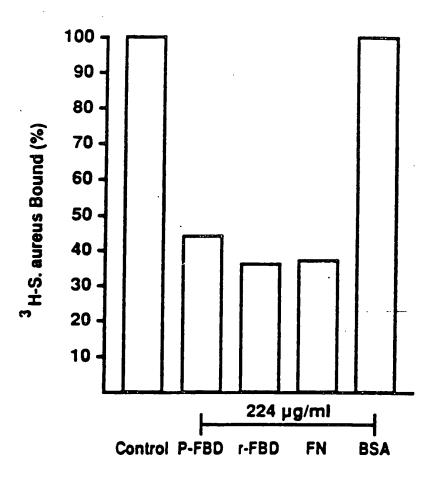
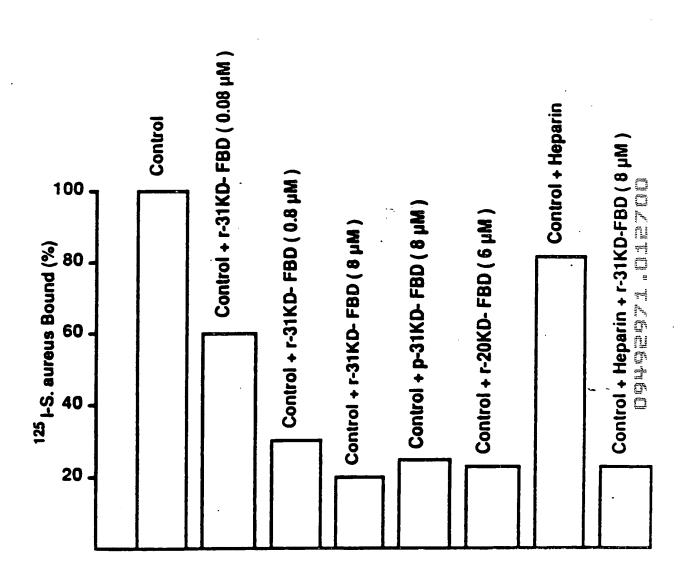


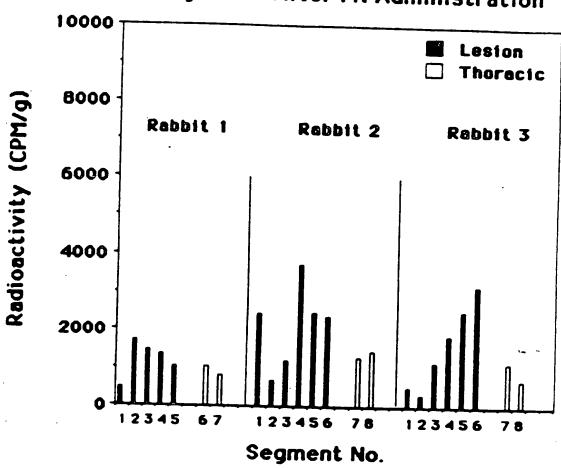
Figure 28

Binding of S. aureus to Bronchial Catheters; Effect of FBD and Heparin.



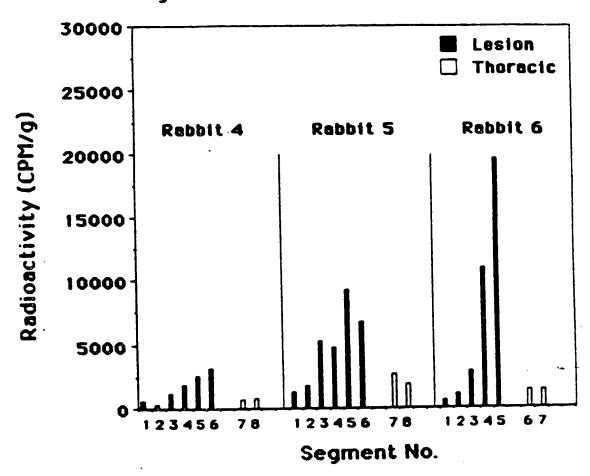
37/62
Figure 29A

Distribution of Radioactivity in Rabbit Aorta Segments After FN Administration



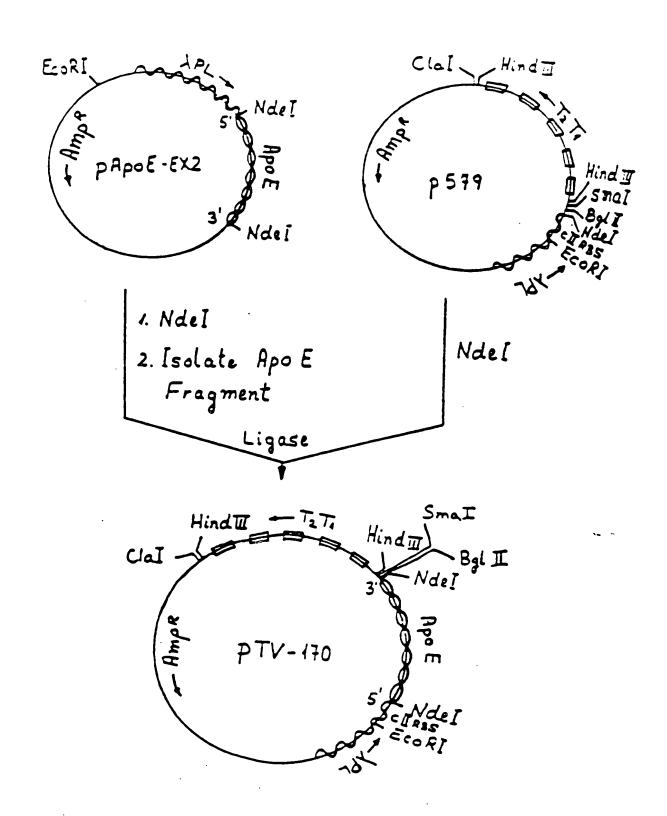
38/62 Figure 29B

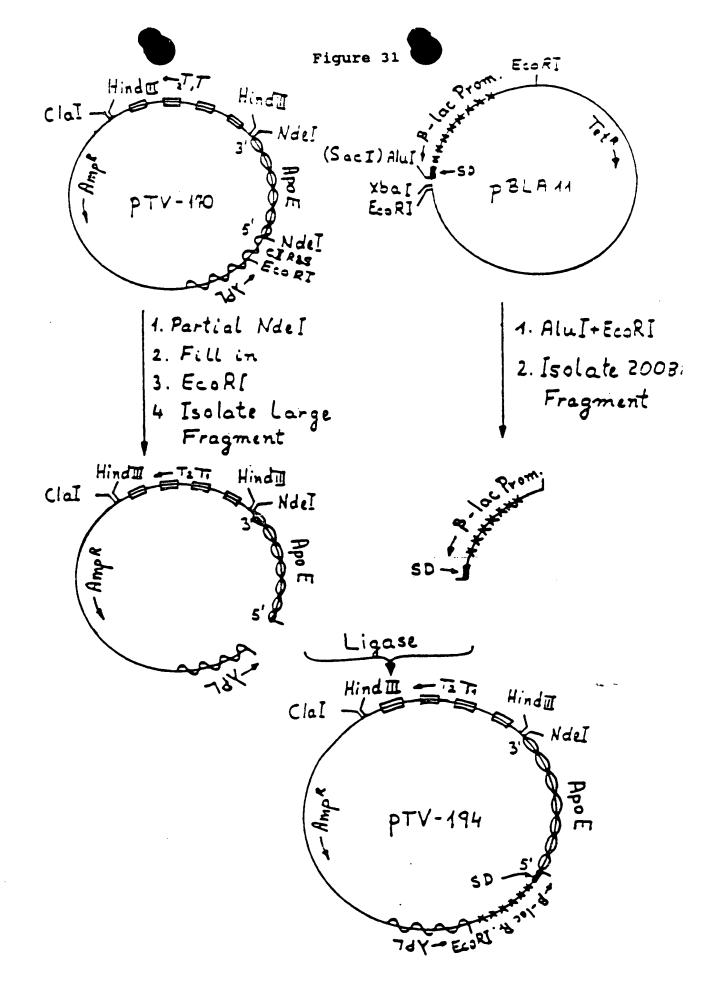
Distribution of Radioactivity in Rabbit Aorta Segments After 31kD FBD Administration

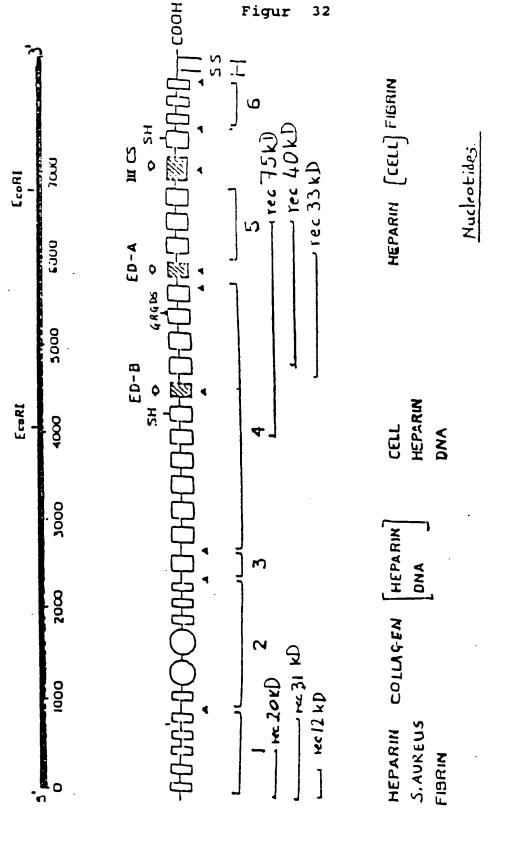


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39/62 Figure 30

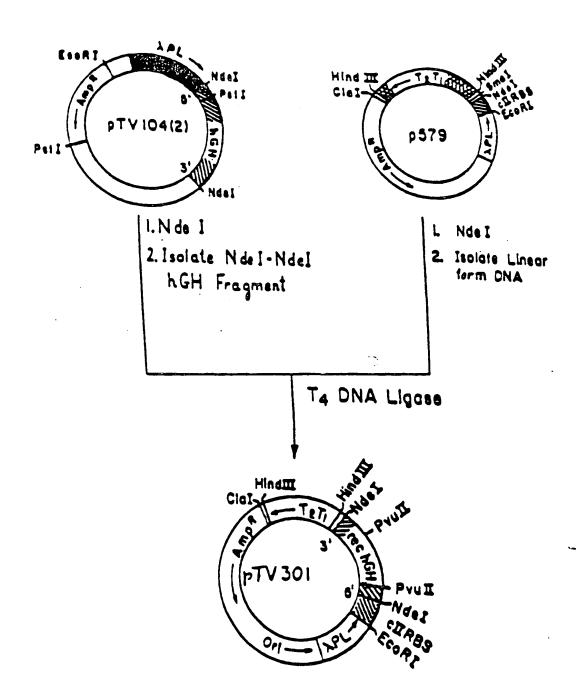


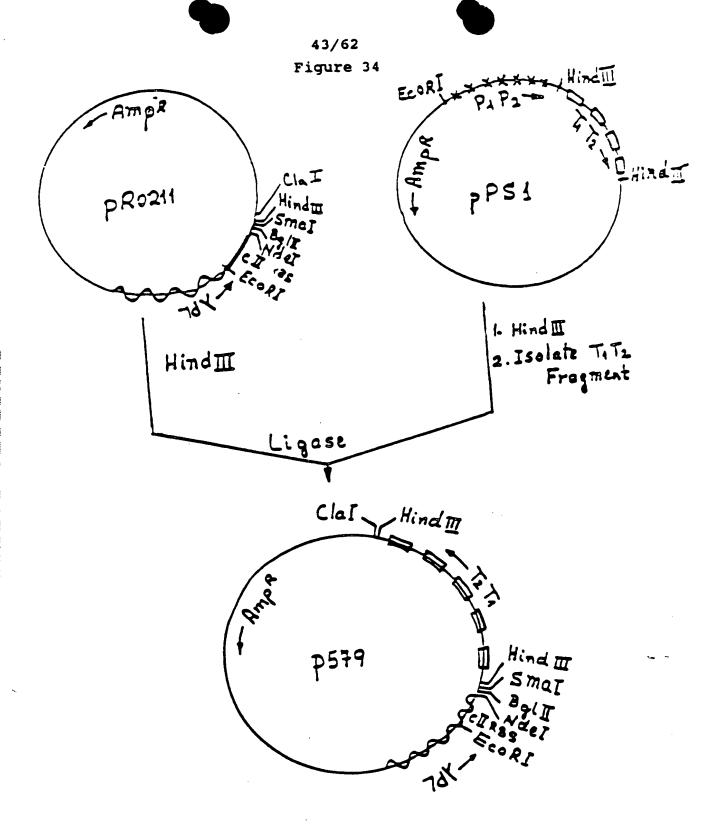


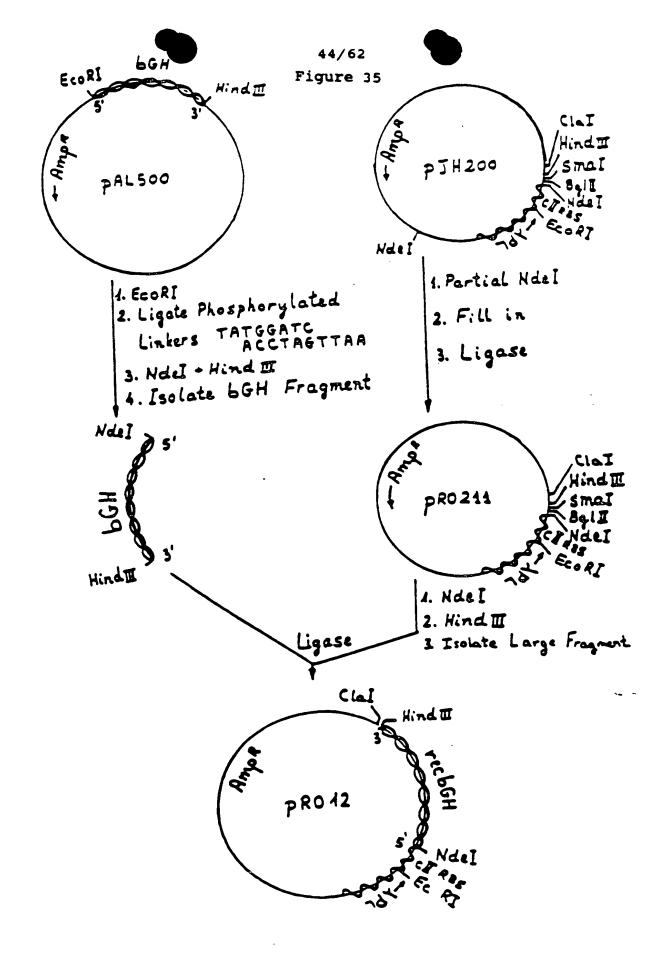


FBD $\frac{11_{\mu} - 340}{\kappa - 20 \text{kD}}$: $\frac{11_{\mu} - 340}{1!_{\mu} - 4!_{\tau}^{2}}$ $\frac{11_{\mu} - 4!_{\tau}^{2}}{1!_{\tau} - 31 \text{kD}}$: $\frac{11_{\mu} - 4!_{\tau}^{2}}{1!_{\tau} - 399}$

42/62 Figur 33



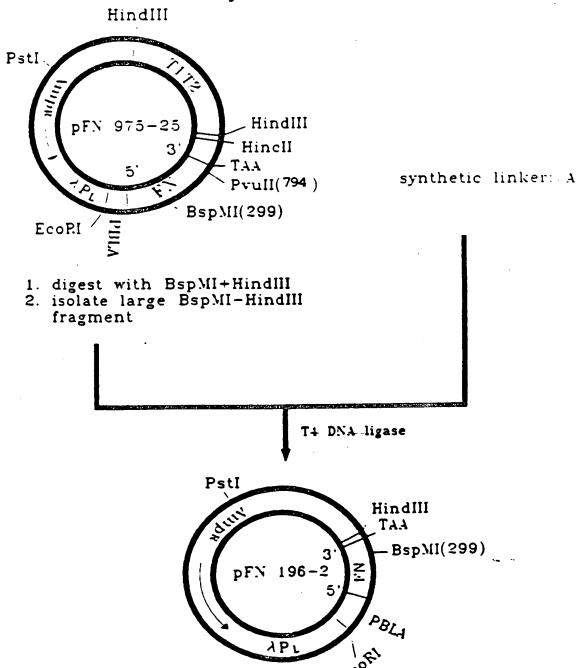




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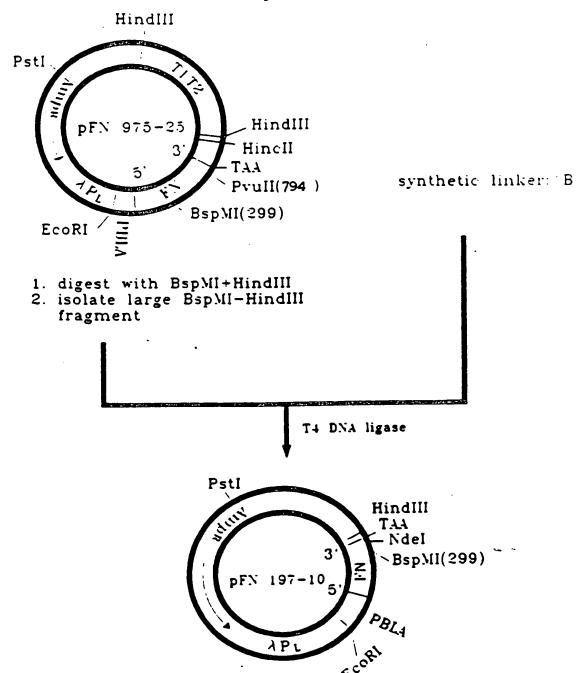
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Figur 36



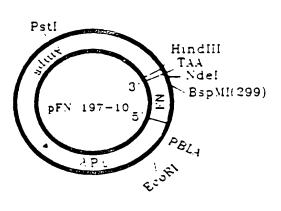
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Figure 37

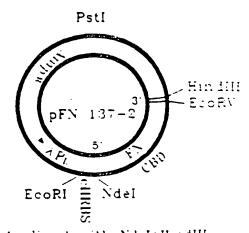


ti,/fbJ

47/62 Figure 38



- 1. digest with Ndel-HindIII 2. isolate large Ndel-HindIII fragment



- digest with Ndel+HindIII
 isolate Ndel-HindIII CBD
- fragment

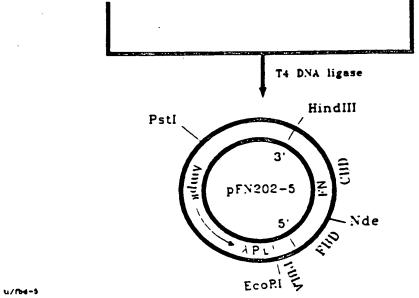
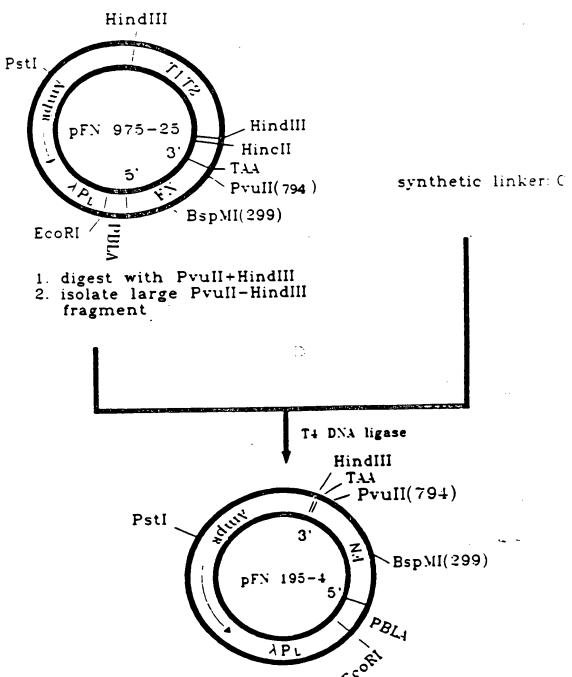
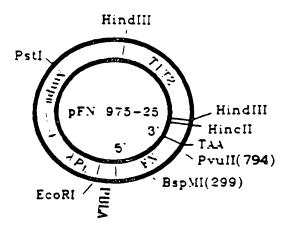


Figure 39

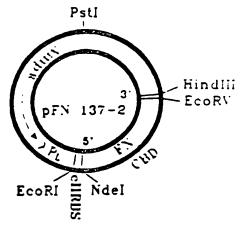


ti/fbd-3

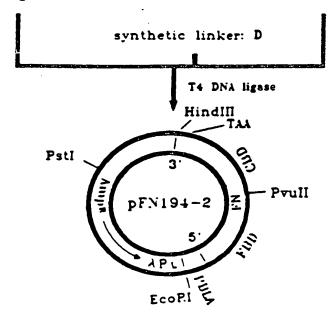
Figure 40



- digest with Pvull+HindIII
 isolate large Pyull-HindIII
- fragment



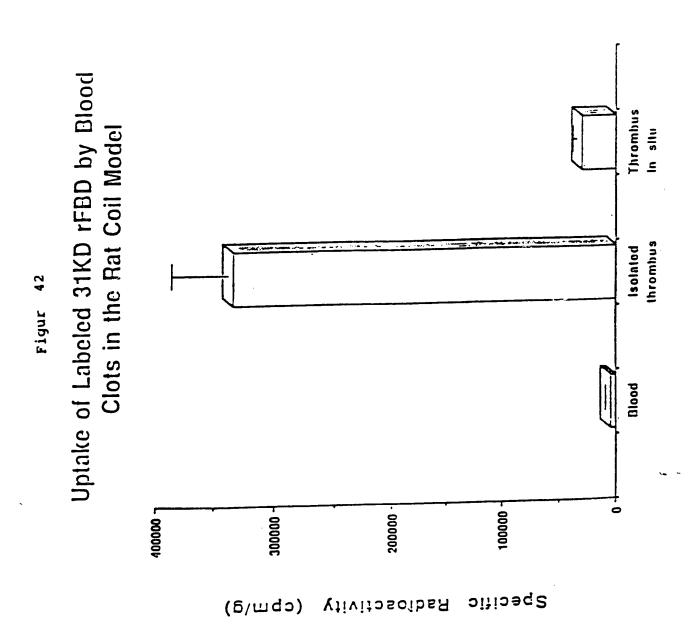
- digest with Ndel+HindIII
 isolate Ndel-HindIII CBD fragment



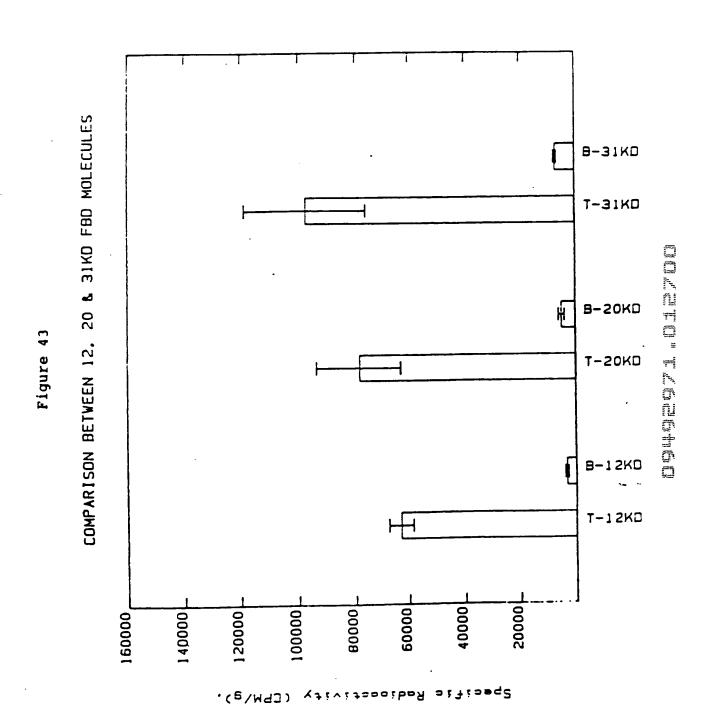
4/04-4

Figure 41

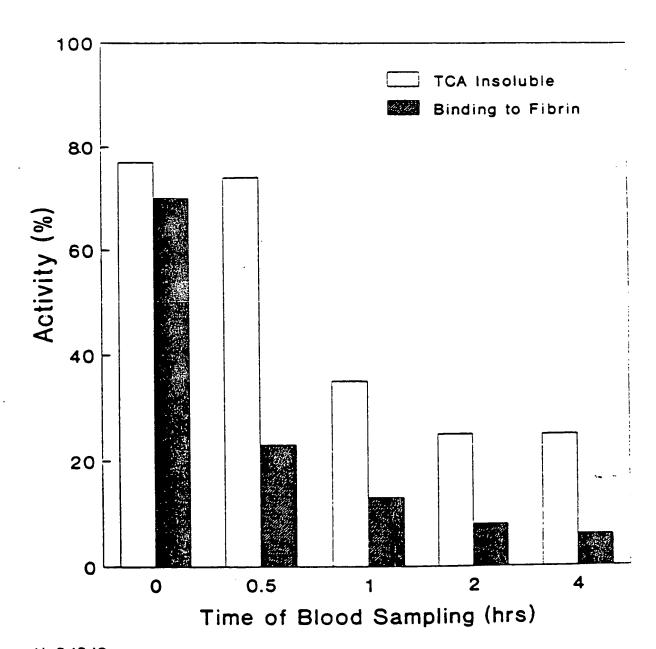
	5 .	GGGCTGGGCGAGGGAG	AATAAGCTGTACCAT	CGCAAACCGCTAACAGCTGA	3
A B	3 .	ACCCGCTCCCTC	TTATTCGACATGGTAC	GCGTTTGGCGATTGTCGACTTCGA	5
	5 ·	GGGCTGGGCGAGGGAG	AATAAGCTGTACCATO	EGCAAACCGCCATATGTAAA	3
	3 .	ACCCGCTCCCTC	TTATTCGACATGGTAG	GCGTTTGGCGGTATACATTTTC&A	5
_	5 ·	ATGGCCGTGGAGACAG	CTAACAGCTGA	3 '	
C	3 '	TACCGGCACCTCTGTCGATTGTCGACTTCGA 5'			
D	5 '	CTGTATACCAACC	3 '		



DAKET TAKES



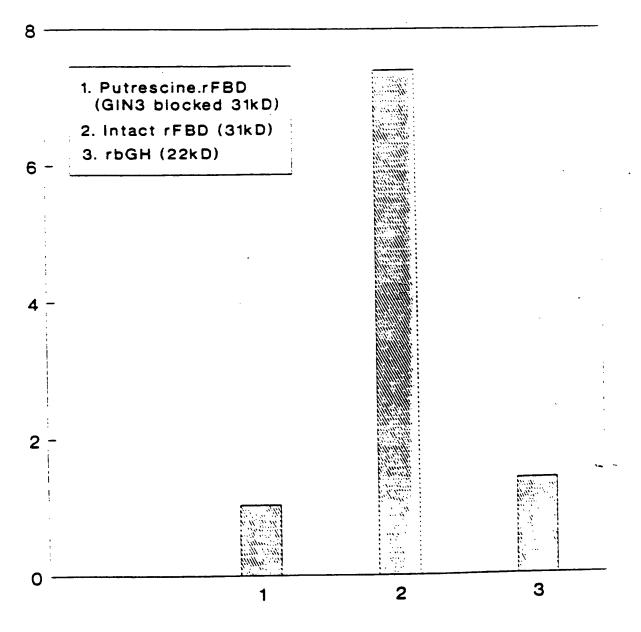
Metabolic Stability of rFBD in Rats; Ex-vivo Binding to Fibrin vs. TCA insolubility



ti-3/6/2

Figure 45

Specificity of binding to Fibrin; Effect of T.G. on the binding of rFBD vs. rbGH (Reaction II)



Figur 46

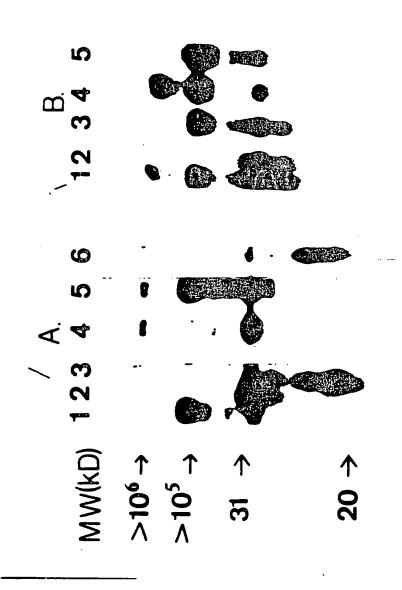
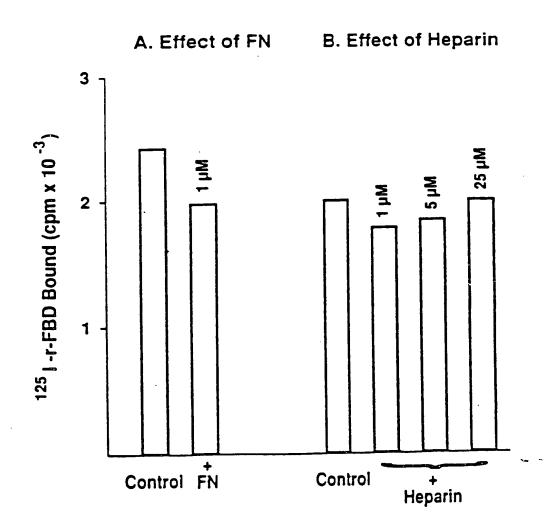
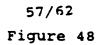




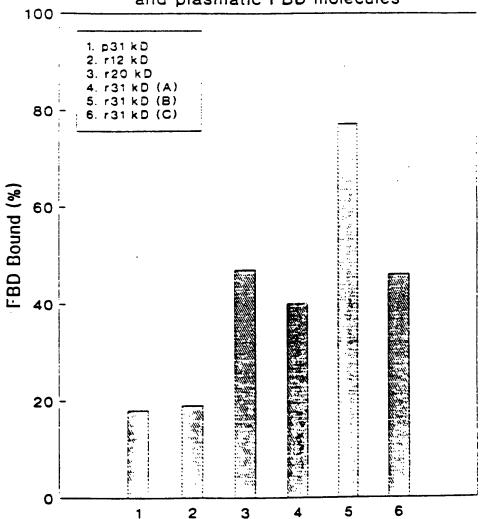
Figure 47

Binding of FBD to preformed clot (Reaction II); Effect of FN and Heparin.



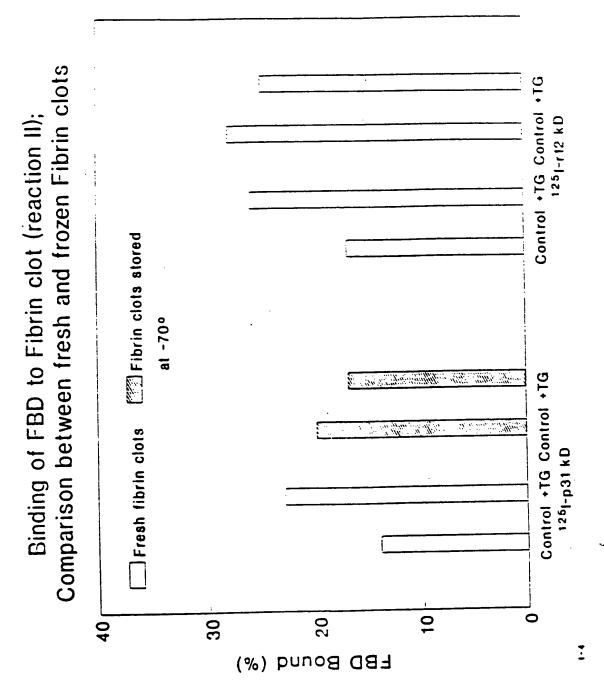


Binding of FBD to Fibrin clot (reaction II); comparison between various recombinant and plasmatic FBD molecules

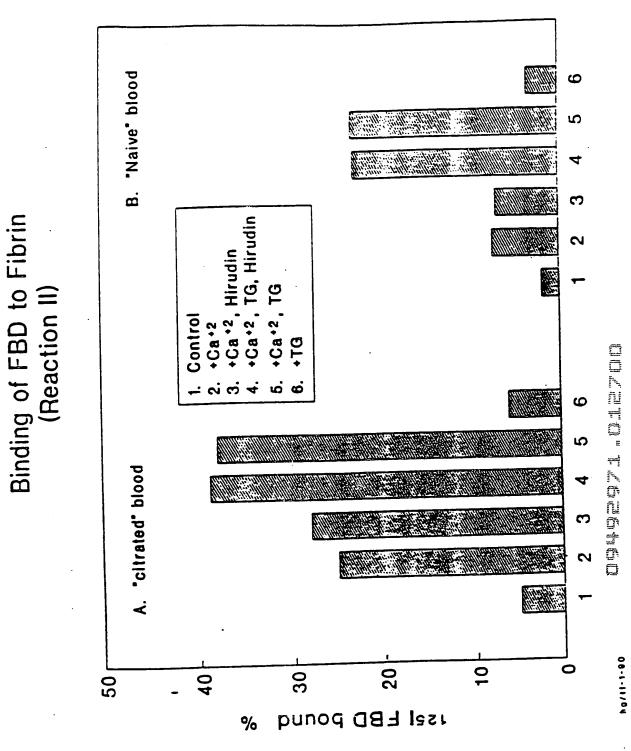


1-3

58/62 Figur 49

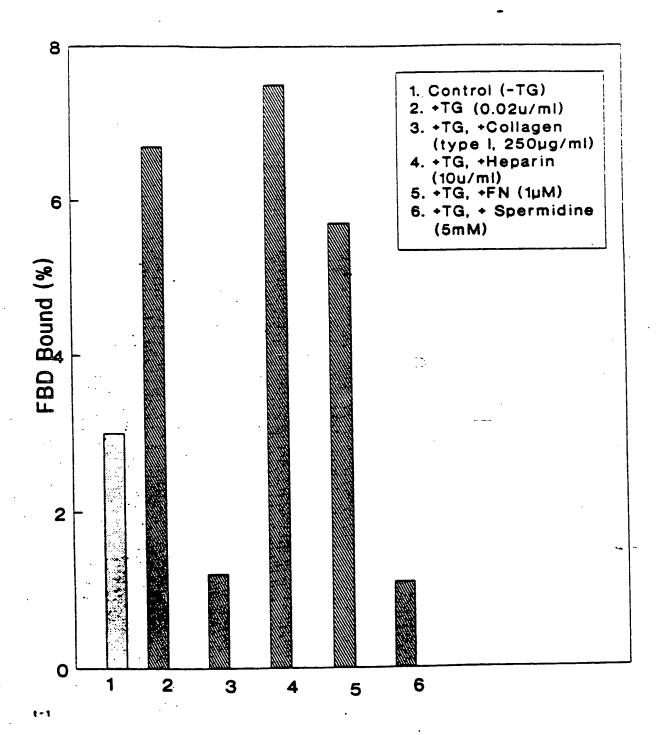


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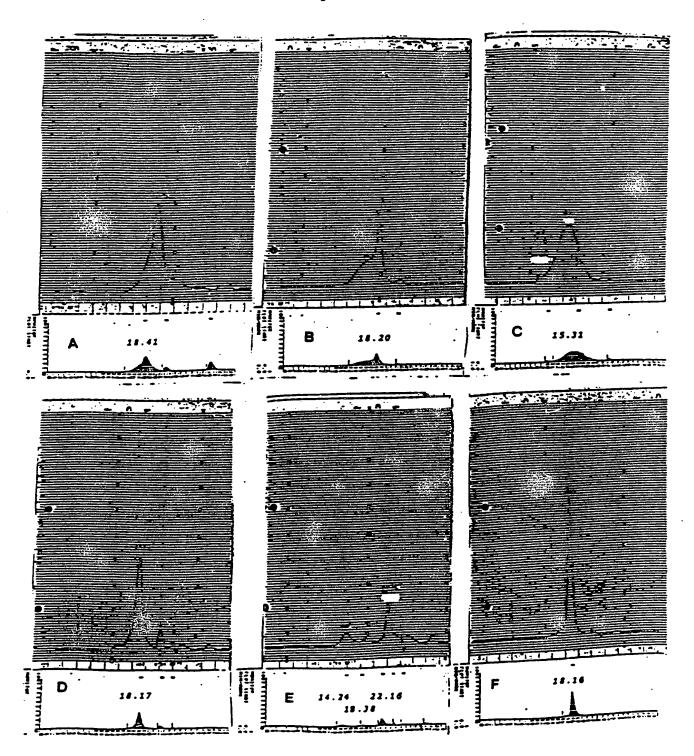


r-FBD binding to ECM



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